Dickmann, Lourdes; Andreotti, Marcelo; Pereira Souza, Marcelo Fernando; Hisashi Nakao, Allan; Christal Catalani, Gabriela

Residual phosphate fertilization and Azospirillum brasilense in the common bean in succession to maize intercropped with Marandu grass

Revista Ciência Agronômica, vol. 48, núm. 3, julio-septiembre, 2017, pp. 404-412
Universidade Federal do Ceará
Ceará, Brasil

Revista Ciência Agronômica, v. 48, n. 3, p. 404-412, jul-set, 2017
Centro de Ciências Agrárias - Universidade Federal do Ceará, Fortaleza, CE
www.ccarevista.ufc.br

Artigo Científico
ISSN 1806-6690

Residual phosphate fertilization and *Azospirillum brasilense* in the common bean in succession to maize intercropped with Marandu grass[1]

Residual da adubação fosfatada e do *Azospirillum brasilense* no feijoeiro em sucessão ao consórcio milho/capim-Marandu

Lourdes Dickmann[2]*, Marcelo Andreotti[2], Marcelo Fernando Pereira Souza[3], Allan Hisashi Nakao[2] and Gabriela Christal Catalani[2]

ABSTRACT - One of the alternatives for achieving sustainable agriculture and a reduction in production costs, especially with phosphate fertilisers, is to inoculate seeds with bacteria of the genus *Azospirillum*. The aim of this study therefore, was to evaluate residual phosphate fertilisation and *Azospirillum brasilense*, together with the contribution of straw from maize intercropped with Marandu grass, on leaf nutritional content, yield components and winter bean yield. The experiment was carried out on the Teaching and Research Farm, of the School of Engineering at UNESP, located in Selvíria in the State of Mato Grosso do Sul, in a typic clayey dystrophic Red Latosol. The experimental design was of randomised blocks with four replications in a 5 x 2 factorial scheme. The treatments consisted of beans sown on straw from maize intercropped with Marandu grass on areas that had received five levels of P_2O_5 in the form of MAP, applied during an initial cultivation of black oats (0, 30, 60, 120 and 240 kg ha^{-1}), both with and without inoculation of the oat and maize which preceded the beans with *Azospirillum brasilense*. Leaf nutrient content, leaf chlorophyll index (ICF), yield components and bean productivity were all evaluated. Inoculation with *Azospirillum brasilense* of the black oat and maize seeds improved the nutritional status of the plants, but had a negative effect on grain yield. Fertilisation of the oat crop with phosphorus had a positive residual effect on the beans, with increases in yield components and grain yield.

Key words: *Urochloa brizantha*. *Phaseolus vulgaris*. Crop-livestock integration. Phosphorus. No-tillage System.

RESUMO - Uma das alternativas para obter agricultura com sustentabilidade e redução dos custos de produção, principalmente com fertilizantes fosfatados, é a inoculação das sementes com bactérias do gênero *Azospirillum*. Assim, o presente estudo teve como objetivo avaliar o residual da adubação fosfatada e do *Azospirillum brasilense* juntamente com a contribuição da palhada do consórcio milho/capim-Marandu sobre os teores foliares nutricionais, componentes de produção e produtividade do feijoeiro de inverno. Conduziu-se o experimento na Fazenda de Ensino, Pesquisa e Extensão da Faculdade de Engenharia/UNESP, localizada em Selvíria/MS, em um Latossolo Vermelho Distrófico típico argiloso. O delineamento experimental foi o de blocos casualizados, com 4 repetições, em esquema fatorial 5 x 2. Os tratamentos foram constituídos da semeadura do feijoeiro sobre a palhada do consórcio milho/capim-Marandu nas áreas das parcelas que receberam as cinco doses de P_2O_5 na forma de MAP aplicadas no cultivo inicial da aveia preta (0; 30; 60; 120 e 240 kg ha^{-1}) e a inoculação ou não com *Azospirillum brasilense* na aveia preta e no milho antecessores ao feijoeiro. Foram avaliados os teores de nutrientes foliares, o índice de clorofila foliar (ICF), os componentes da produção e produtividade do feijoeiro. A inoculação das sementes de aveia preta e do milho com *Azospirillum brasilense* em consórcio com capim-Marandu, antecessores ao cultivo do feijoeiro, melhorou o estado nutricional das plantas, mas exerceu efeito negativo na produtividade de grãos. A adubação fosfatada da cultura da aveia proporcionou efeito residual positivo para o feijoeiro com incremento dos componentes da produção e produtividade de grãos.

Palavras-chave: *Urochloa brizantha*. *Phaseolus vulgaris*. Integração lavoura-pecuária. Fósforo. Sistema plantio direto.

DOI: 10.5935/1806-6690.20170047
*Autor para correspondência
Recebido para publicação em 26/01/2016; aprovado em 11/09/2016
[1]Parte da Dissertação de Mestrado financiada pela Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP)
[2]Departamento de Fitossanidade, Engenharia Rural e Solos, Universidade Estadual Paulista "Júlio de Mesquita Filho"/UNESP, Campus de Ilha Solteira-SP, Brasil, lourdesdickmann@hotmail.com, dreotti@agr.feis.unesp.br, allannakao@hotmail.com, gchristalcatalani@yahoo.com.br
[3]Secretaria de Ciência de Estado e Tecnologia/Escola Técnica Estadual de Alta Floresta-MT, celonando@hotmail.com

INTRODUCTION

The common bean (*Phaseolus vulgaris*), the staple food of millions of Brazilians (MELÉM JUNIOR *et al*., 2011), is actually a legume of the utmost importance and high social significance, with a winter production of 1.222 kg ha^{-1} in 662,000 ha (CONAB, 2015).

In cultivated areas, the common bean is one of the main annual species adaptable to No Tillage System (NTS), with high productivity and great relevance in aspersion-irrigation systems during the intercrop period (FARINELLI; LEMOS, 2010). The bean crop is greatly promising due to its flexibility in the sowing period and to its short vegetal cycle (REICHERT, 2012).

In low altitude savannah regions, the agriculture-livestock integration by the intercropping of two types of grasses improves grain and forage production during the summer and later in straw formation for the sowing of winter crops (BORGHI; CRUSCIOL, 2007). Fertilized tropical forages increase the production of dry matter and nutrients by organic matter, with a later enhancement by beans in succession (FIORENTIN *et al*., 2012).

Straw coverage demonstrates its initial importance in soil and water conservation and, in the long run, it improves chemical, physical and biological fertility conditions (VILELA *et al*., 2011), enhancing stability to the system and increase in the crop´s grain productivity. (CHIODEROLI *et al*., 2012).

Phosphorus is one of the basic elements in the plants´ early life when they require its availability in the soil. Low use of the element by crops in Brazil affects productivity and, consequently, plant nutrition (GAZOLA *et al*., 2013). Due to Fe and Al oxides in savannah soil, which is mainly latosol, phosphorus is fast adsorbed to these oxides and its mobility and availability to crops are highly reduced. In fact, it becomes a limiting factor in production (VALDERRAMA *et al*., 2011).

Combining the plants´ genetic potential and the soil´s biological resources, such as diazotrophic bacteria, particularly those of the genus *Azospirillum*, is an alternative for the reduction of mineral fertilizers and for obtaining production increase at lower costs and without damaging the environment (NOVAKOWISKI *et al*., 2011).

Diazotrophic bacteria do not only fix atmospheric N_2 but they enhance growth and the root´s absorption surface, benefitting increase in worn soil with their basic role in the absorption of slow nutrients in the soil, such as phosphorus (OKON; VANDERLEYDEN, 1997). Cangahuala-Inocente *et al*. (2013) report that *Azospirillum* is active in the synthesis of auxins, cytokinins, gibberellins, ethylene and other molecules, and in the solubilization of phosphates in the soil.

There is a high demand in agribusiness for low cost technologies, high productivity and reduction in environmental impacts (GONÇALVES *et al*., 2013). Current research evaluates residual phosphate fertilization and inoculation with *Azospirillum brasilense* on nutritional foliar rates, production components and bean winter crop productivity in succession to maize-Miranda grass intercropping.

MATERIALS AND METHODS

The assay was developed during the 2014 harvest on the Teaching, Research and Extension Farm of the Engineering Faculty of Ilha Solteira/Universidade Estadual Paulista “Júlio de Mesquita Filho” (FE/UNESP) in the municipality of Selvíria MS Brazil (20°20’05’’ S and 51°24’26’’ W, altitude 335 m). According to Köppen´s classification, climate in Selvíria MS Brazil is Aw, characterized as humid tropical climate with a rainy season during the summer and a dry season in the winter. Daily data on maximum, mean and minimum temperatures, rainfall and relative air humidity were collected at the meteorological station on the farm, during the experiment (Figure 1).

According to the Brazilian System for Soil Classification, the soil of the experimental area is dystrophic clayey Red Latosol (EMBRAPA, 2013). The area has been NTS cultivated for the last eleven years with annual crops. Previous cultures featured bean (harvest 2014) in current research, maize-Marandu grass intercropping (harvest 2013-2014) and black oats (harvest 2013), as in the sequence: black oats - maize intercropped with *U. brizantha* cv. Marandu and bean winter crop.

In the case of previous crops (maize intercropped with *U. brizantha* cv. Marandu and black oats), treatments comprised five doses of P_2O_5 (0; 30; 60; 120 and 240 kg ha^{-1} P_2O_5) applied only at the sowing of black oats and the inoculation or not with *Azospirillum brasilense* of black oats and maize-Marandu grass intercropping.

The chemical characterization of the soil prior to the sowing of the bean crop followed Raij *et al*. (2001). Ten simple samples per plot were collected by screw auger at a depth between 0 and 0.20 m. Compound samples were produced for chemical analysis (Table 1).

Experimental design comprised randomized 5 x 2 blocks with four replications, or rather, five residual doses of P_2O_5 (0; 30; 60; 120 and 240 kg ha^{-1} P_2O_5) applied

Figure 1 - Data on climate collected at the meteorological station of FEPE during the experiment

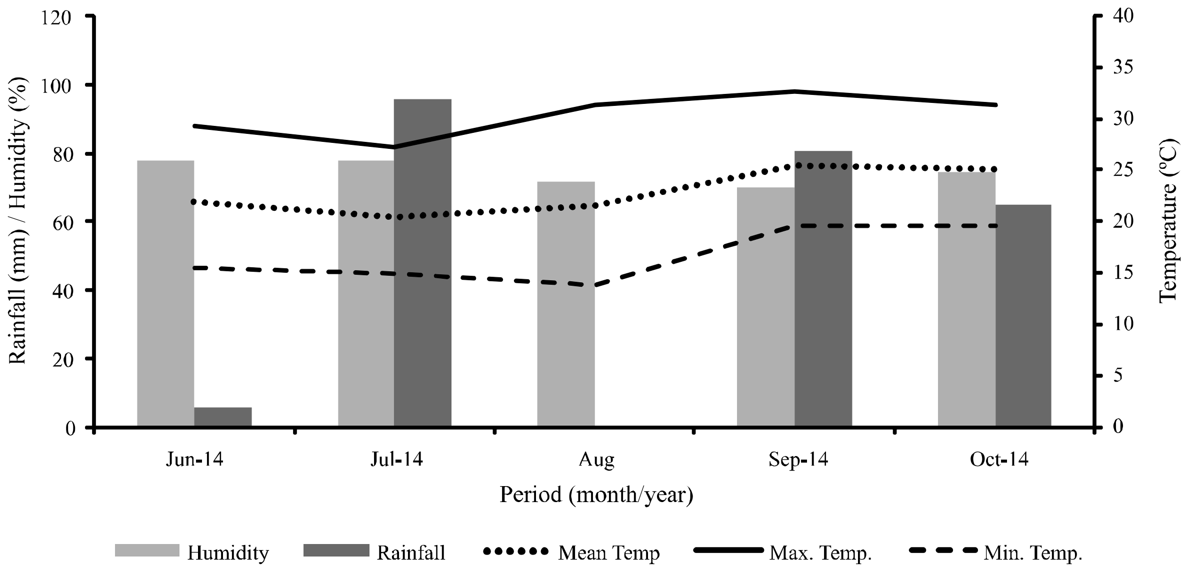


Table 1 - Fertility rate of soil at 0 – 0.20 m layer prior to the sowing of bean crop, due to inoculation or not with *Azospirillum brasilense* and residual phosphate fertilization

Attribute	P_{resin}	MO	pH	K	Ca	Mg	H+Al	SB	CTC	S (mg m^{3})	V (%)
	(mg dm^{-})	(g dm^{-3})	($CaCl_2$)	------------------ (mmol$_c$ dm^{-3}) -----------------							
P_2O_5 doses					without inoculation						
0	15.0	18.0	5.4	2.2	21.0	18.0	26.0	40.7	68.9	4.0	58.0
30	14.0	18.0	5.6	1.6	28.0	22.0	25.0	49.6	73.6	5.0	67.0
60	13.0	19.0	5.4	1.6	27.0	21.0	26.0	48.9	76.1	5.0	63.0
120	21.0	19.0	5.5	1.3	26.0	20.0	26.0	48.3	74.3	6.0	65.0
240	19.0	19.0	5.6	1.9	25.0	19.0	25.0	45.1	69.6	6.0	65.0
P_2O_5 doses					with inoculation						
0	13.0	19.0	5.3	1.9	21.0	20.0	28.0	42.1	70.4	6.0	60.0
30	22.0	19.0	5.5	1.8	26.0	24.0	26.0	49.8	77.8	5.0	66.0
60	22.0	19.0	5.6	2.1	26.0	22.0	25.0	49.1	73.6	5.0	67.0
120	16.0	19.0	5.4	1.1	25.0	20.0	26.0	46.3	71.8	6.0	64.0
240	17.0	18.0	5.3	1.5	23.0	22.0	27.0	46.2	73.5	6.0	63.0

during the sowing of black oats in June 2013, with or without inoculation in black oat and maize (maize + *U. brizantha* cv. Marandu) seeds with diazotrophic bacteria *Azospirillum brasilense* provided by Total AZO inoculant at the commercial dose 100 mL 25 kg^{-1} seeds. Seed inoculation occurred in the shade immediately before sowing. Experimental area measured 1,760 m^{2}, with plots 4.4 m wide and 10 m long, totaling 44 m^{2}.

Bean sowing occurred on the straw coverage of maize-*U. brizantha* cv. Marandu intercropping and the areas were inoculated or not with *Azospirillum brasilense* in black oats and in maize-*U. brizantha* cv. Marandu, antecessors of the bean crop. The five phosphorus doses were applied in the black oat crop previous to the culture of maize-*U. brizantha* cv. Marandu and bean crop.

Marandu grass was desiccated by the herbicide Glyphosate at a dose of 1.440 g ha^{-1} of active ingredient prior to bean sowing, whilst plants were ground by a vegetal residue horizontal grinder (Triton).

Bean seeds cv. Pérola were mechanically sown by a sowing-fertilizing machine locked to a machete-type groover for NTS, with a 0.45 m space between rows and a distribution of 15 seeds per m^{-1}. Further, 50 g of Carboxina + 50 g of Tiran/100 kg of seeds were used for seed treatment.

Seeding fertilizer comprised 20 kg of N and 40 kg ha^{-1} of K_2O, with urea and KCl as sources, respectively. Cover fertilization occurred on the 30th day after plant emergence (stage V4) with 50 kg ha^{-1} of N as urea.

The Foliar Chlorophyll Index (FCI) and foliar nutrient rates were calculated during the flowering of the bean crop (V4-R5). Digital chlorophyll-meter (CFL 1030 - Falker) was employed at the third totally developed trifoliate leaf to determine FCI, with an average of 10 readings per leaflet in ten plants per plot. Twenty mature trifoliate leaves (3rd trifoliate) per plot were harvested on the same day when foliar FCI readings were performed. They were dried in a forced air buffer at 65 ºC till constant mass and ground in a Wiley mill to determine macronutrient rates, following methodology by Malavolta, Vitti and Oliveira (1997).

The morphological attributes, production components and grain productivity were calculated at the end of the crop cycle. Plants of the usable area (four 2m-long rows at the center) in each plot were harvested and assessed as to the number of plants per experimental unit to determine the plant´s final situation (PFS), the height of the insertion of the first pod (HIFP) in 10 plants per plot.

Since ten plants harvested at random in the usable area per plot determined production components, the number of pods per plant (NPP), the number of grains per pod (NGP) and the mass of 100 grains (M100) could be assessed. Results were corrected for 13% humidity. All the plants of the plot´s usable area were mechanically threshed and weighed to evaluate grain productivity (GP); humidity was corrected at 13% (wet base) and the final result was transformed at kg ha^{-1}. Correction of grain humidity was required due to differences in maturation within the same harvest and to avoid the development of microorganisms (fungus and insects).

Shapiro-Wilk´s test (1965), under nil hypothesis of normality, was employed to test normality at 5% significance. Results underwent analysis of variance by F-test ($p<0.05$). Effect of inoculation or non-inoculation with *Azospirillum brasilense* in antecessors black oats and maize were compared by Tukey´s test ($p < 0.05$). Effect of residual phosphate fertilization was assessed by regression analysis. Statistical analyses were performed with SISVAR® (FERREIRA, 2011).

RESULTS AND DISCUSSION

Data underwent normality test and the nil hypothesis was not rejected, or rather, residues showed normal distribution at 5% significance.

The inoculation with *A. brasilense* of intercropping maize antecessor to bean crop altered P, Ca and Mg rates available in the bean´s leaf tissues. Residual phosphate fertilization and the combination of treatments did not affect nutrition rates and FCI of the bean crop (Table 2).

When *A. brasilense* was employed in intercropping maize antecessor to the bean crop, results were not significant as the rates of foliar P of the bean crop, with greater rates (5.2 g kg^{-1}) with bean crops in non-inoculated areas (Table 2). However, regardless of inoculation or not by the diazotrophic bacterium, mean rates for P in bean plants were above the culture´s adequate levels (between 2 and 3 g kg^{-1} P), according to Malavolta, Vitti and Oliveira (1997).

When Balzergue *et al*. (2011) applied higher doses of P_2O_5 in peas during the early phase of colonization by shrubby mycorrhizal fungi (SMF), the authors reported the complete disappearance of the symbiosis, perhaps due to self-regulation, partially as a response to signalization by plants. When plants are cultivated in high phosphate concentrations, they neither estimate SMF nor do they present strigolactone, with a decrease or lack of radicular colonization in the wake of high phosphate doses. Consequently, absorption is reduced.

Ca and Mg rates in bean leaves were higher in plants from plots in which maize intercropped with Marandu grass was inoculated through seeds (12.5 and 5.1 g kg^{-1}, respectively). According to Hungria *et al*. (2010), bacteria of the genus *Azospirillum* have a fundamental role in the enhancement of plant growth and may improve the absorption of several macro- and micronutrients by enhancing the efficiency of nutrients available in the soil.

According to Raij *et al*. (1997), the adequate range lies between 10 and 25 g kg^{-1} and 2.5 and 5.0 g kg^{-1} of dry matter for Ca and Mg, respectively. Results reveal that management of bean winter crop intercropped with maize-Marandu grass with maize inoculation by *A. brasilense* was highly relevant for Ca absorption since non-inoculated plant rates, albeit adequate for the crops, are at the lowest limits of sufficiency, according to Raij *et al*. (1997).

It should be enhanced that high concentrations of Mg, verified in current analysis, may cause a nutritional imbalance in leaf tissues, with consequences for crop development (RAIJ *et al*., 1997). Therefore, *A. brasilense* may raise Mg levels in leaf tissues above the culture´s critical line and cause damage in the tissues due to nutrient

Table 2 - Average Foliar Chlorophyll Index (FCI) and rates of foliar macronutrients in bean winter crop due to inoculation or not by *Azospirillum brasilense* in maize intercropped with *U. brizantha* cv. Marandu and residual phosphate fertilization

Treatments	FCI	N	P	K	Ca	Mg	S
		g kg^{-1}					
				Inoculation – I			
Without	40.95 a	49.6 a	5.2 a	25.8 a	11.0 b	4.9 b	2.6 a
With	43.15 a	51.5 a	4.9 b	26.4 a	12.5 a	5.1 a	2.5 a
F test	0.58^{ns}	1.47^{ns}	4.29*	0.48^{ns}	9.84**	5.51*	0.21^{ns}
SMD	5.95	3.33	0.23	1.76	0.98	0.14	0.20
				Doses of P_2O_5 – D			
0	42.49	48.0	5.1	24.8	11.8	5.1	2.7
30	37.49	48.1	5.0	27.3	11.4	4.9	2.4
60	46.99	52.9	5.0	25.5	12.3	5.1	2.7
120	38.63	50.7	5.3	26.5	11.6	4.9	2.5
240	44.66	53.1	5.0	26.5	12.0	5.0	2.5
F Test – D	1.52^{ns}	1.84^{ns}	0.67^{ns}	1.02^{ns}	0.38^{ns}	0.64^{ns}	1.21^{ns}
SMD	13.39	7.50	0.52	3.99	2.21	0.31	0.45
F Test – I x D	1.38^{ns}	1.66^{ns}	2.02^{ns}	0.38^{ns}	0.38^{ns}	0.36^{ns}	0.65^{ns}
CV (%)	6.46	10.15	7.08	10.49	12.83	4.32	12.26

Averages followed by the same letter in the column do not differ by Tukey´s test at 5% probability. ** and *: significant at 1 and 5% probability by F Test, respectively. ^{ns} not significant. SMD: significant minimum difference; CV: coefficient of variation

excess. Results for Mg may also be related to the good levels of the nutrient available at the soil´s cultivable layer (0 – 0.20 m), as Table 1 shows, enhancing greater absorption and redistribution of the nutrient in the plant due to the bacteria. When *A. brasilense* is employed for Mg nutrition in bean crops, the Mg levels already in the soil should be taken into account since they are prone to rise when bacteria are present and attain the crop´s critical levels.

N rates in the bean plant´s foliar tissues did not increase by applying *A. brasilense*, perhaps due to nitrogenated fertilization in sowing and coverage. It is a well-known fact that the biological fixation of nitrogen decreases when mineral N is provided. Consequently, the mineral fertilization of N in the crop may have caused the non-occurrence of positive effects of the nutrient in foliar tissues, as Table 2 shows.

Several factors contribute towards the complexity of inoculation responses with diazotrophic bacteria, including the interaction of plant genotype and inoculated strain, the plants´ nutritional conditions, soil and the environment´s abiotic factors (LEMOS *et al*., 2013; QUADROS *et al*., 2014).

The residual effect of phosphate fertilization in the sowing of black oats failed to affect the macronutrient rates of the bean´s foliar tissues (Table 2). Lack of significant results in the increase of foliar rates may be due to the nutrient´s low mobility in the soil and to its application prior to the sowing of black oats. According to Santos, Gatiboni and Kamiski (2008), under such conditions, the greatest P accumulations occur at the soil´s surface layers in the No Tilling System due to the consecutive addition of fertilizers on the surface layer, lack of soil turning and decrease in erosion rates. In fact, the above are characteristics similar to the soil in the studied area, with more than 10 years of NTS, as Table 1 shows.

There was a significant difference between treatments with and without inoculation of intercropping maize antecessor to the bean crop. The application of *A. brasilense* provided lower productivity rates of the bean crop when compared to that in non-inoculated plants (Table 3). Results were similar to those by Campos, Theisen and Gnatta (2000), since no response on grain yield was available when they studied the use of the inoculant Graminante®, perhaps due to the bacteria specific to the assessed genotype.

However, bean productivity rates in current research are above the Brazilian average and within the average for the state of São Paulo, regardless of the plants´ inoculation by *A. brasilense* (CONAB, 2015).

Table 3 - Average number of plants (NP), number of pods per plant (NPP), number of grains per plant (NGP), number of grains per pod (NGPD), height of insertion of the first pod (HIFP), height of plant (HP), mass of 100 grains and productivity (PROD) of the bean winter crop due to the inoculation or non-inoculation of maize intercropped with *U. brizantha* cv. Marandu with *Azospirillum brasilense* and residual phosphate fertilization

Treatments	NP	NPP	NGP	NGPP	HIFP	HP	M100 G	PROD kg ha^{-1}
	---------- n° ----------				--- cm ---			
				Inoculation – I				
without	210.556 a	17.2 a	89.5 a	5.2 a	19.76 a	69.21 a	24.52 a	2.874 a
with	215.833 a	15.9 a	84.4 a	5.3 a	20.10 a	69.74 a	25.16 a	2.625 b
F test	0.37^{ns}	3.98^{ns}	1.76^{ns}	0.63^{ns}	0.11^{ns}	0.10^{ns}	1.53^{ns}	16.97**
SMD	17.041	1.40	7.90	0.21	1.23	3.57	1.10	123.66
				Doses of P_2O_5 – D				
0	212.500	15.8	79.5	5.0	19.90	68.10	24.75	2.551
30	202.778	16.1	84.8	5.3	19.08	68.68	25.03	2.637
60	205.556	15.9	84.6	5.3	20.08	68.01	25.30	2.779
120	220.833	18.3	99.5	5.6	20.25	72.63	24.76	2.843
240	224.306	16.7	86.5	5.2	20.50	69.95	24.36	2.937
F test – D	0.93^{ns}	1.75^{ns}	3.03*	3.54*	0.65^{ns}	0.99^{ns}	0.37^{ns}	5.35**
Model equation	-	-	Q	Q	-	-	-	L
R^2 (%)	-	-	78.67	93.40	-	-	-	85.71
F test - E	-	-	7.44*	12.34**	-	-	-	-
F test – I x D	0.22^{ns}	1.82^{ns}	3.71^{ns}	2.74^{ns}	0.66^{ns}	0.48^{ns}	0.62^{ns}	18.35**
CV (%)	12.85	13.06	14.00	6.07	9.50	7.92	6.53	10.89

Averages followed by the same letter in the column do not differ by Tukey´s test at 5% probability. ** and *: significant at 1 and 5% probability by F Test, respectively. ns not significant. SMD: significant minimum difference; CV: coefficient of variation

No significant difference between treatments with or without inoculation of maize antecessor to the bean crop has been reported for the production components, probably due to the non-specificity of diazotrophic bacteria from the soil to the bean crop. This fact may have decreased the infection rate of bean roots by the bacteria of the genus *Rhizobium*.

The lack of significant responses to treatments may be also due to the good fertility of the soil of the experimental area, including N, which may have reduced the multiplication of *A. brasilense* and thus enhanced the absence of responses for the assessed agronomic characteristics and production components.

The remaining P_2O_5 doses altered the number of grains per plant and the number of grains per pod (Table 3), whereas the interaction between the remaining P_2O_5 doses and inoculation or non-inoculation of maize antecessor to the bean crop by *A. brasilense* had a significant effect on bean productivity (Table 4).

Plants from plots in which maize was not inoculated and associated to areas with applications 30 and 60 kg ha^{-1} P_2O_5 and without residual P (control), revealed bean productivity which was greater than that in plots with inoculated maize (Table 4). The above was probably due to the effect described by Balzergue *et al*. (2011). In other words, plants cultivated in high phosphate concentrations are unable to stimulate SMF and do not have strigolactone, with absorption reduction and a consequent decrease or lack of radicular colonization due to high phosphate doses.

Productivity results of inoculated areas provided a positive linear adjustment to the increase of the soil´s residual P_2O_5 (Table 4) by reduction of rates in the soil after the exportation of antecessor maize grains and by the lack of SMF participation. On the other hand, there was no significant regression adjustment in areas in which the antecessor intercropping maize was not inoculated with *A. brasilense*, evidenced by NTS in the area (nutrient cycling|) and by adequate P rates in the soil, even in the control plot (Table 1).

Table 4 - Development of the interaction inoculation with *Azospirillum brasilense* of maize intercropped with *U. brizantha* cv. Marandu and residual phosphate fertilization for the productivity of bean winter plant

Inoculation	P_2O_5 (kg ha^{-1})					Equation	R^2 (%)
	0	30	60	120	240		
	Productivity (kg ha^{-1})						
without	2,762 a	2,868 a	3,159 a	2,768 a	2,811 a	-	ns
with	2,339 b	2,405 b	2,399 b	2,919 a	3,064 a	Y= 2.324,3916+3,3423x**	87.47

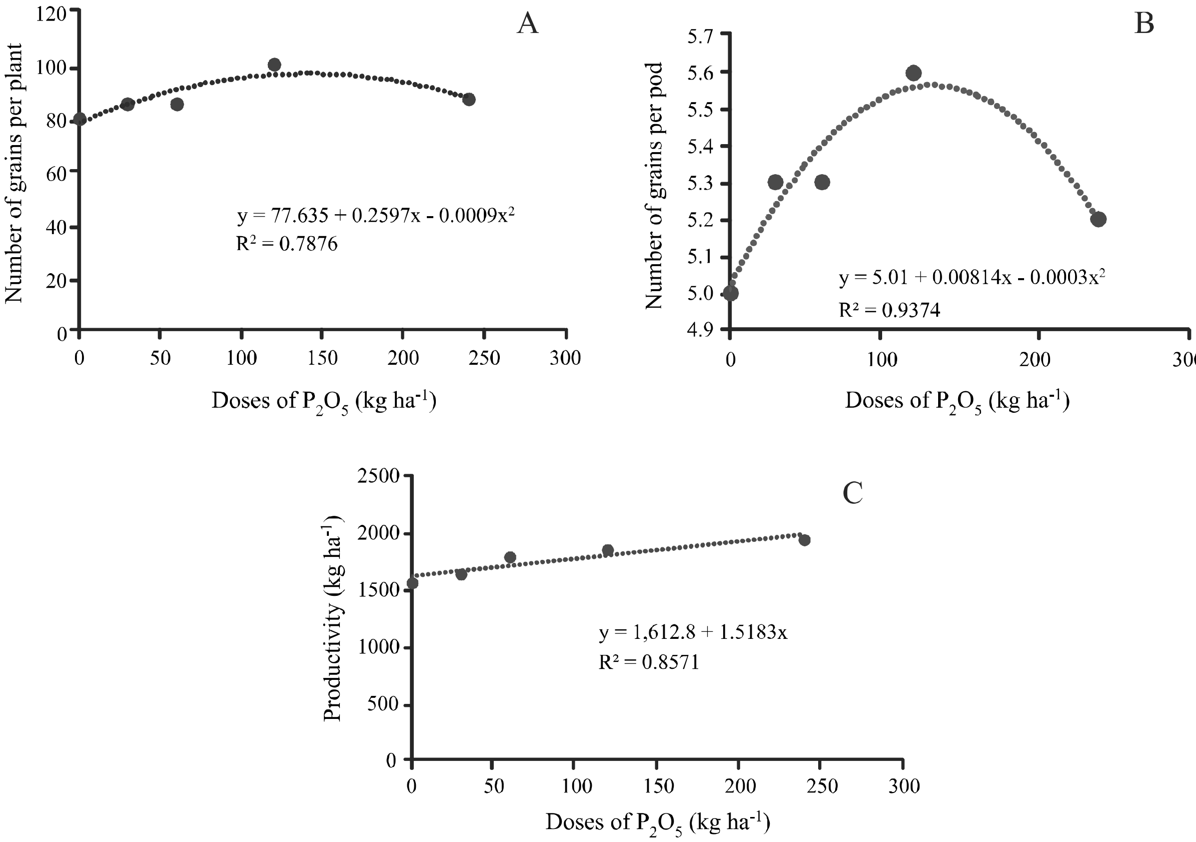
Averages followed by the same letter in the column do not differ by Tukey´s test at 5% probability. ** and *: significant at 1 and 5% probability, respectively. ns not significant

Results on the number of grains per plant and per pod caused by P_2O_5 doses adjusted to a quadratic model (Figures 2A and 2B) with maximum peaks, respectively, for doses 144 and 136 kg ha^{-1} of P_2O_5. Similarly to the number of grains per plant (Figure 2A) and to the number of grains per pod (Figure 2B) and, probably, due to the same, the bean plant´s productivity responded positively to P_2O_5 doses in the sowing of black oats and with residual effect for the bean winter plant a year after fertilization.

Grain productivity results proved to be linear (Figure 2C), corroborating data by Nascente *et al*. (2014) who reported increase in productivity of bean plants when doses of P_2O_5 increased.

Santos *et al*. (2011) also underscored the importance of phosphate fertilization on bean production, even though P is the least absorbed among macronutrients due to the characteristics of high soil adsorption and to the fact that the nutrient is only slightly available to vegetation.

Figure 2 - Number of grains per plant (A), number of grains per pod (B) and productivity of bean winter plant (C) due to residual phosphate fertilization



According to Raij (2001), adequate phosphorus doses are an asset for the development of the crop from the start to the end of its cycle, stimulating radicular growth, formation of reproduction parts and fruit formation. Adequate P levels in the soil provide plants with greater production and with more resistance against adversities (ZUCARELI *et al*., 2011). It is highly probable that during the plants´ growth and development, P2O5 doses which cause the highest production rates supplemented the bean´s nutritional requirements with equilibrium.

CONCLUSIONS

1. Inoculation with *Azospirillum brasilense* in seeds of black oats and maize intercropped with Marandu grass anteceding the bean winter crop improved the plants´ nutritional state by a linear increase in grain productivity only in areas in which the anteceding plants were inoculated by *A.* brasilense;
2. There was an increase in production components caused by the residual effect of phosphate fertilization, with maximum peaks between 144 and 136 kg ha^{-1} of P_2O_5, respectively for the number of grains per plant and the number of grains per pod.

ACKNOWLEDGEMENTS

The authors would like to thank the Foundation Research Support of the State of São Paulo (FAPESP) for the scholarship awarded to the first author (Process FAPESP n. 2013/03439-3) and for funding current research.

REFERENCES

BALZERGUE, C. *et al*. The regulation of arbuscular mycorrhizal symbiosis by phosphate in pea involves early and systemic signaling events. **Journal of Experimental Botany**, v. 62, p. 1049-1060, 2011.

BORGHI, E.; CRUSCIOL, C. A. C. Produtividade de milho, espaçamento e modalidade de consorciação com *Brachiaria brizantha* em sistema plantio direto. **Pesquisa Agropecuária Brasileira**, v. 42, n. 2, p. 163-171, 2007.

CAMPOS, B. H. C.; THEISEN, S.; GNATTA, V. Avaliação de inoculante graminante na cultura do milho. **Ciência Rural**, v. 30, n. 4, p. 713-715, 2000.

CANGAHUALA-INOCENTE, G. C. *et al.* Identification of six differentially accumulated proteins of *Zea mays* seedlings (DKB 240 variety) inoculated with *Azospirillum brasilense* strain FP2. **European Journal of Soil Biology**, v. 58, p. 45-50, 2013.

CHIODEROLI, C. A. *et al*. Atributos físicos do solo e produtividade de soja em sistema de consórcio milho e braquiária. **Revista Brasileira de Engenharia Agrícola e Ambiental**, v. 16, n. 1, p. 37-43, 2012.

COMPANHIA NACIONAL DE ABASTECIMENTO. **Acompanhamento de safra brasileira**: grãos, primeiro levantamento, outubro, 2015. Disponível em: <http://www.conab.gov.br/OlalaCMS/uploads/arquivos/15_10_09_09_03_07_boletim_graos_outubro_2015.pdf.>. Acesso em: 15 jan. 2015.

EMPRESA BRASILEIRA DE PESQUISA AGROPECUÁRIA. Centro Nacional de Pesquisa de Solos. **Sistema Brasileiro de Classificação de Solos**. 3. ed. Brasília, DF , 2013. 353 p.

FARINELLI, R.; LEMOS, L. B. Produtividade, eficiência agronômica, características nutricionais e tecnológicas do feijão adubado com nitrogênio em plantio direto e convencional. **Bragantia**, v. 69, n. 1, p.165-172, 2010.

FERREIRA, D. F. Sisvar: a computer statistical analysis system. **Ciência e Agrotecnologia**, v. 35, n. 6, p. 1039-1042, 2011.

FIORENTIN, C. F. *et al*. Adubação nitrogenada de cobertura no feijoeiro de inverno-primavera em três sistemas de cultivo. **Semina: Ciências Agrárias**, v. 33, p. 2825-2836, 2012.

GAZOLA, R. N. *et al.* Efeito residual da aplicação de fosfato monoamônio revestido por diferentes polímeros na cultura de milho. **Revista Ceres**, v. 60, n. 6, p. 876-884, 2013.

GONÇALVES, M. V. *et al.* Efeito do composto orgânico, biofertilizante e inoculante no rendimento de feijão-caupi vigna unguiculata (L.) walp. *In*: CONGRESSO NACIONAL DE FEIJÃO-CAUPI, 3., 2013, Recife. **Anais eletrônicos...** Recife: CONAC, 2013. Disponível em: < http://www.conac2012.org/resumos/pdf/191a.pdf>. Acesso em: 13 jan. 2016.

HUNGRIA, M. *et al*. Inoculation with selected strains of *Azospirillum brasilense* and *A. lipoferum* improves yields of maize and wheat in Brazil. **Plant and Soil**, v. 331, n. 1/2, p. 413-425, 2010.

LEMOS, J. M. *et al*. Resposta de cultivares de trigo à inoculação de sementes com *Azospirillum brasilense*, e à adubação nitrogenada em cobertura. **Científica**, v. 41, n. 2, p. 189-198, 2013.

MALAVOLTA, E.; VITTI, G. C.; OLIVEIRA, S. A. **Avaliação do estado nutricional das plantas**: princípios e aplicações. 2. ed. Piracicaba: Associação Brasileira para Pesquisa da Potassa e do Fosfato, 1997. 319 p.

MELÉM JÚNIOR, N. J. M. *et al.* Nutrição mineral e produção de feijão em áreas manejadas com e sem queima de resíduos orgânicos e diferentes tipos de adubação. **Semina: Ciências Agrárias**, v. 32, n. 1, p. 7-18, 2011.

NASCENTE, A. S. *et al*. Adubação fosfatada no sulco e foliar afetando a produtividade de grãos do feijoeiro comum. **Semina: Ciências Agrárias**, v. 35, n. 3, p. 1231-1240, 2014.

NOVAKOWISKI, J. H. *et al.* Efeito residual da adubação nitrogenada e inoculação de *Azospirillum brasilense* na cultura do milho. **Semina: Ciências Agrárias**, v. 32, n. 4, p. 1687-1698, 2011.

OKON, Y.; VANDERLEYDEN, J. Root-associated *Azospirillum* species can stimulate plants. **Applied and Environmental Microbiology**, v. 63, n. 7, p. 366-370, 1997.

QUADROS, P. D. *et al.* Desempenho agronômico a campo de híbridos de milho inoculados com *Azospirillum*. **Revista Ceres**, v. 61, n. 2, p. 209-218, 2014.

RAIJ, B. van. *et al.* **Análise química para avaliação da fertilidade de solos tropicais**. Campinas: Instituto Agronômico, 2001. 284 p.

RAIJ, B. van. *et al.* **Recomendações de adubação e calagem para o estado de São Paulo**. Campinas: Instituto Agronômico Campinas, 1997. 285 p.

REICHERT, P. **Cultura do feijão e nitrogênio no sistema de integração lavoura-pecuária**. 2012. 54 f. Dissertação (Mestrado em Agronomia) - Curso de Pós-graduação em Agronomia, Universidade Estadual do Centro-Oeste, Guarapuava, 2012.

SANTOS, D. R.; GATIBONI, L. C.; KAMISKI, J. Fatores que afetam a disponibilidade do fósforo e o manejo da adubação fosfatada em solos sob sistema plantio direto. **Ciência Rural**, v. 38, n. 2, p. 576-586, 2008.

SANTOS, J. Z. L. *et al.* Resposta do feijoeiro à adubação fosfatada em solos de cerrado com diferentes históricos de uso. **Revista Brasileira de Ciência do Solo**, v. 35, p. 193-202, 2011.

SHAPIRO, S. S.; WILK, M. B. An analysis of variance test for normality (complete samples). **Biometrika**, v. 52, n. 3/4, p. 591-611, 1965. Disponível em: <http://biomet.oxfordjournals.org/content/52/3-4/591.full.pdf+html>. Acesso em: 23 mar. 2016.

VALDERRAMA, M. *et al*. Fontes e doses de NPK em milho irrigado sob plantio direto. **Pesquisa Agropecuária Tropical**, v. 41, n. 2, p. 254-263, 2011.

VILELA, L. *et al*. Sistemas de integração lavoura-pecuaria na região do Cerrado. **Pesquisa Agropecuária Brasileira**, v. 46, n. 10, p. 1127-1138, 2011.

ZUCARELI, C. *et al.* Fósforo na produtividade e qualidade de sementes de feijão Carioca Precoce cultivado no período das águas. **Revista Ciência Agronômica**, v. 42, n. 1, p. 32-38, 2011.